Claims

1. A method for identifying a nucleic acid molecule which encodes a catalytic protein, said method comprising the steps of:

a) providing a candidate catalytic protein fusion molecule, comprising a candidate catalytic protein linked to both its nucleic acid coding sequence and a substrate; and

b) determining whether said candidate catalytic protein catalyzes a reaction of said substrate by assaying for an alteration in molecular size, charge, or conformation of said fusion molecule, relative to an unreacted fusion molecule, thereby identifying a nucleic acid molecule which encodes a catalytic protein.

2. The method of claim 1, wherein said alteration in molecular size, charge, or conformation of said reacted fusion molecule is detected by an alteration in electrophoretic mobility.

3. The method of claim 1, wherein said alteration in molecular size, charge, or conformation of said reacted fusion molecule is detected by column chromatography.

4. The method of claim 3, wherein said alteration in molecular size, charge, or conformation of said reacted fusion molecule is detected by HPLC, FPLC, ion exchange column chromatography, or size exclusion chromatography.

5. A method for identifying a nucleic acid molecule which encodes a catalytic protein, said method comprising the steps of:

a) providing a candidate catalytic protein fusion molecule, comprising a candidate catalytic protein linked to both its nucleic acid coding sequence and a substrate;

b) allowing said candidate catalytic protein to catalyze a reaction of said substrate in solution;

c) contacting the product of step (b) with a capture molecule that has specificity for and binds a reacted fusion molecule, but not an unreacted fusion molecule, said capture molecule being immobilized on a solid support; and

d) detecting said reacted fusion molecule in association with said solid support,

thereby identifying a nucleic acid molecule which encodes a catalytic protein.

6. The method of claim 6, wherein, as a result of said reaction, said substrate is covalently bonded to an affinity tag and said capture molecule binds said affinity tag but does not bind an unreacted fusion molecule.

7. A method for identifying a nucleic acid molecule which encodes a catalytic protein, said method comprising the steps of:

a) providing a candidate catalytic protein fusion molecule, comprising a candidate catalytic protein linked to both its nucleic acid coding sequence and a substrate, said substrate being covalently bonded to an affinity tag;

b) allowing said candidate catalytic protein to catalyze a reaction of said substrate in solution;

c) contacting the product of step (b) with a capture molecule that is specific for said affinity tag, said capture molecule being immobilized on a solid support; and

d) determining whether said fusion molecule is bound to said solid support, wherein the determination that a fusion molecule is not bound to said solid support identifies a nucleic acid molecule which encodes a catalytic protein.

8. The method of claim 7, wherein said solid support is a column or beads and a fusion molecule that does not bind to said column includes a nucleic acid molecule which encodes a catalytic protein.

9. A method for identifying a nucleic acid molecule which encodes a catalytic protein, said method comprising the steps of:

a) providing a candidate catalytic protein fusion molecule, comprising a candidate catalytic protein linked to both its nucleic acid coding sequence and a substrate;

b) allowing said candidate catalytic protein to catalyze a reaction of said substrate in solution in the presence of an affinity tag, said reaction resulting in the covalent attachment of said affinity tag to said fusion molecule;

c) immunoprecipitating the product of step (b) with an antibody that is specific for said affinity tag; and

d) detecting said immunoprecipitation complex, thereby identifying said fusion molecule as having a nucleic acid molecule which encodes a catalytic protein.

10. The method of claim 1, 5, 7, or 9, wherein said candidate catalytic protein fusion molecule is present in a population of candidate catalytic protein fusion molecules.

11. The method of claim 1, 5, 7, or 9, wherein said substrate is a protein.

12. The method of claim 1, 5, 7, or 9, wherein said substrate is a nucleic acid.

13. The method of claim 12, wherein said nucleic acid is RNA.

14. The method of claim 1 or 7, wherein said catalytic protein is a ribonuclease and said substrate is RNA.

15. The method of claim 1, 5, or 9, wherein said catalytic protein is an RNA ligase, an RNA polymerase, a terminal transferase, a reverse transcriptase, or a tRNA synthetase and said substrate is RNA.

16. The method of claim 12, wherein nucleic acid is DNA.

17. The method of claim 1 or 7, wherein said catalytic protein is a deoxyribonuclease or a restriction endonuclease and said substrate is DNA.

18. The method of claim 1, 5, or 9, wherein said catalytic protein is a DNA ligase, a terminal transferase, a DNA polymerase, or a polynucleotide kinase and said substrate is DNA.

19. The method of claim 1, 5, or 9, wherein said substrate is covalently bonded to said candidate catalytic protein fusion molecule.

20. The method of claim 7 or 19, wherein said substrate is a substrate-nucleic acid conjugate and the nucleic acid portion of said conjugate is linked to the nucleic acid portion of said candidate catalytic protein fusion molecule.

21. The method of claim 7 or 19, wherein said substrate is a protein and is linked to the protein portion of said candidate catalytic protein fusion molecule.

22. The method of claim 1, 5, or 9, wherein said substrate is non-covalently associated with said candidate catalytic protein fusion molecule.

23. The method of claim 22, wherein said substrate is covalently bonded to a nucleic acid strand hybridized to the nucleic acid portion of said candidate catalytic fusion molecule.

24. The method of claim 1, 5, 7, or 9, wherein said nucleic acid coding sequence of said candidate catalytic protein fusion molecule is double-stranded.

25. The method of claim 1, wherein, in step (b), said determining step is carried out by assaying for an alteration in molecular size, charge, or conformation of the nucleic acid coding sequence of a fragment thereof.

26. The method of claim 5, wherein, in step (d), said detecting step is carried out by detecting the nucleic acid coding sequence or a fragment thereof in association with said solid support.

27. The method of claim 7, wherein, in step (d), said determining step is carried out by determining whether or not the nucleic acid coding sequence or a fragment thereof is bound to said solid support.

28. The method of claim 9, wherein, in step (d), said detecting step is carried out by detecting the nucleic acid coding sequence or a fragment thereof in said immunoprecipitation complex.

29. A method for identifying a nucleic acid molecule which encodes an autoproteolytic protein, said method comprising the steps of:

a) providing a candidate autoproteolytic protein fusion molecule, comprising a candidate autoproteolytic protein linked to its nucleic acid coding sequence; and

b) determining whether said candidate autoproteolytic protein catalyzes a self-reaction by assaying for an alteration in molecular size, charge, or conformation of said fusion molecule, relative to an unreacted fusion molecule, thereby identifying a nucleic acid molecule which encodes an autoproteolytic protein.

30. The method of claim 29, wherein said alteration in molecular size, charge, or conformation of said reacted fusion molecule is detected by an alteration in electrophoretic mobility.

31. The method of claim 29, wherein said alteration in molecular size, charge, or conformation of said reacted fusion molecule is detected by column chromatography.

32. The method of claim 31, wherein said alteration in molecular size, charge, or conformation of said reacted fusion molecule is detected by HPLC, FPLC, ion exchange column chromatography, or size exclusion chromatography.

33. A method for identifying a nucleic acid molecule which encodes an autoproteolytic protein, said method comprising the steps of:

a) providing a candidate autoproteolytic protein fusion molecule, comprising a candidate autoproteolytic protein linked to its nucleic acid coding sequence;

b) allowing said candidate autoproteolytic protein to self-react;

c) contacting the product of step (b) with a capture molecule that has specificity for and binds a self-reacted fusion molecule, but not an unreacted fusion molecule, said capture molecule being immobilized on a solid support; and

d) detecting said self-reacted fusion molecule in association with said solid support, thereby identifying a nucleic acid molecule which encodes an autoproteolytic protein.

34. A method for identifying a nucleic acid molecule which encodes an autoproteolytic protein, said method comprising the steps of:

a) providing a candidate autoproteolytic protein fusion molecule, comprising a candidate autoproteolytic protein linked to its nucleic acid coding sequence, said protein being covalently bonded to an affinity tag;

b) allowing said candidate autoproteolytic protein to self-react in solution;

c) contacting the product of step (b) with a capture molecule that is

specific for said affinity tag, said capture molecule being immobilized on a solid support; and

d) determining whether said fusion molecule is bound to said solid support, wherein the determination that a fusion molecule not bound to said solid support identifies a nucleic acid molecule which encodes an autoproteolytic protein.

35. The method of claim 34, wherein said solid support is a column or beads and a fusion molecule that does not bind to said column includes a nucleic acid molecule which encodes an autoproteolytic protein.

36. A method for identifying a nucleic acid molecule which encodes an autoproteolytic protein, said method comprising the steps of:

a) providing a candidate autoproteolytic protein fusion molecule, comprising a candidate autoproteolytic protein linked to its nucleic acid coding sequence;

b) allowing said candidate autocatalytic protein to self-react in solution;

c) immunoprecipitating the product of step (b) with an antibody that is specific for a reacted fusion molecule; and

d) detecting said immunoprecipitation complex, thereby identifying said fusion molecule as having a nucleic acid molecule which encodes an autoproteolytic protein.

37. The method of claim 29, 33, 34, or 36, wherein said candidate autoproteolytic protein fusion molecule is present in a population of candidate autoproteolytic protein fusion molecules.

38. The method of claim 29, 33, 34, or 36, wherein said autoproteolytic protein is a self-cleaving enzyme.

39. The method of claim 29, 33, 34 or 36, wherein said autoproteolytic protein is a self-splicing enzyme.

40. The method of claim 29, 33, 34, or 36, wherein said nucleic acid coding sequence of said candidate autoproteolytic protein fusion molecule is double-stranded.